REMARKS

Reconsideration and allowance are respectfully requested.

Claims 1-13 and 24 have been examined. Claims 14-23 were withdrawn from consideration by the Examiner. Claims 19-23 are amended. Claim 25 is added.

The amendments are supported by the original disclosure and, thus, no new matter has been added. If the Examiner should disagree, however, she is respectfully requested to point out the challenged limitation with particularity in the next Action so support may be cited in response.

Unity of Invention/Election

The special technical feature shared by the compounds recited in claim 1 is that they are all Schiff base forming compounds. The compounds can be patentably distinct from each other and still share a special technical feature. If so, the compounds of the claim would be directed to a single general inventive concept.

Finally, Applicants have not stated that they regard each compound as a distinct invention (cf. last sentence of first paragraph on page 3 of the Action) because whether or not the recited Schiff base forming compounds are distinct inventions is not relevant to concluding that they share a special technical feature.

New claim 25 should be examined as directed to the elected invention.

Examination of vaccine compositions (i.e., claims 14-17) and methods of making compositions of components (i.e., claims 18-23) is requested in view of the examination of the product claim 24.

35 U.S.C. 103 – Nonobviousness

Claims 1-9, 11-13 and 24 were rejected under Section 103(a) as allegedly being unpatentable over Rhodes (US 5,508,310) in view of Herrmann et al. (US 5,620,896). Moreover, claim 10 was rejected under Section 103(a) as allegedly being unpatentable over Rhodes in view of Herrmann et al. and Bellhouse et al. (US 5,620,896). Applicants traverse both rejections and request reconsideration.

The evidence of record discloses that tucaresol (an example of the Schiff base forming compounds recited in claim 1) is an adjuvant for conventional protein vaccines. At issue is whether the prior art demonstrates that one of ordinary skill in the art would have been motivated to make the combination/modification proposed by the Examiner and a reasonable expectation of success. Applicants submit that the claimed invention is not prima facie obvious because neither motivation nor reasonable expectation of success is shown by the evidence of record: the different mechanisms and objectives of protein vaccines and DNA vaccines do not support the use of a Schiff base forming compound as an adjuvant for DNA vaccines based on its use as an adjuvant for protein vaccines. Furthermore, none of the cited references teaches or suggests that tucaresol would "enhance both humoral and cellular immune responses initiated by the antigenic peptide" in the context of DNA vaccination as required by claims 1-24.

Applicants can refute absolutely the assertion that the mechanism of tucaresol's mechanism of action is the same regardless of whether it is used as an adjuvant for a conventional protein vaccine or a DNA vaccine according to Applicants' invention. These two mechanisms are not the same. Moreover, at the time the invention was made, there were good reasons to have believed that mechanistically tucaresol would not work in the setting of a DNA vaccine.

Rhodes defined **immunopotentiator** as "an agent which is capable of restoring a depressed immune function, or enhancing normal immune function, or both" (column 1 lines 52-57). Subsequently, **immune function** was defined as "the development and expression of humoral (antibody-mediated) immunity, cellular (T-cell-mediated) immunity, or macrophage and granulocyte mediated resistance" (column 5, lines 24-28). This term was being used in the context of so-called wild-type infections and other disease settings such as malignancy, where the antigens to which the immune response is to be enhanced are provided by tumors in and infections of the body (see column 1, lines 7-12). Rhodes also provides for the use of tucaresol as an adjuvant for a protein vaccine (see column 14, lines 33-37). He suggests that a vaccine may be prepared by formulating the antigenic component with the compound. Rhodes used tucaresol in a setting in which the protein antigens to which the immune response was

to be enhanced would be co-administered with the adjuvant, i.e., as a conventional protein vaccine.

Neither Rhodes nor Herrmann et al. was contemplating the use of tucaresol (or any other Schiff base forming compound) to enhance the immune response to a DNA vaccine. For the reasons set out below, the two cases are quite different. In both cases (immunopotentiation and vaccine adjuvancy), tucaresol was shown to be exerting its effects by amplifying co-stimulatory or 2nd signals of the immune response. The mechanism of handling both wild-type antigens contributed by an infection and conventional vaccine antigens (see ref. 1 of Appendix II) differs from the way DNA-encoded antigens are handled in a number of fundamental ways. In an infection, antigens are generated by: pathway A in which the synthetic machinery of the host cell when it is taken over by the virus for the production of viral proteins (see attached Fig. A of Appendix I) or pathway B in which whole micro-organisms or fragments of micro-organisms taken up from the inter-cellular environment by phagocytic cells (see attached Fig. B of Appendix I).

The same two routes operate for conventional vaccines in which pathway A predominates for live attenuated vaccines and pathway B predominates for killed and sub-unit vaccines. Importantly, both pathways provide an array of danger signals and co-stimulatory signals initiated by pathogen associated molecular patterns (PAMPs) during the uptake/entry phase into APC (refs. 2-4 of Appendix II).

In contrast, antigens provided by DNA encoded vaccines are taken up in a different way and utilize a unique mechanism of antigen handling that involves neither of these pathways. Importantly, DNA vaccines do not contribute the array of danger/co-stimulatory signals initiated by PAMPs and other microbial elements. This is illustrated in Fig. C of Appendix II which shows that plasmid DNA directly transfects the cell and lacks the microbial elements that amplify co-stimulation (ref. 5 of Appendix II).

Subsequent events in terms of binding to MHC molecules and ligating the T-cell receptor are the same for all three kinds of delivery (ref. 1 of Appendix II). However, the important difference is in the co-stimulatory environment which is absent in DNA vaccination as shown in pathway C (see attached Fig. C of Appendix II). Adjuvants exert their effects on the co-stimulatory environment (ref. 6 of Appendix II) and both conventional

adjuvants and tucaresol are effective in pathways A and B. It is therefore surprising that tucaresol was found to work in the case of DNA vaccination (i.e., pathway C) where the co-stimulatory environment is absent or very weak.

Adjuvants work on co-stimulatory mechanisms and do not affect the recognition of antigen by the T-cell receptor or the signal it transduces (refs. 6-7 of Appendix II and see Fig. D of Appendix I). Instead, they work through ancillary receptors such as toll-like receptors to amplify co-stimulation of (ref. 8 of Appendix II). Accessory/co-stimulatory signals are known to be interdependent and integrated at a number of levels within antigen presenting cells and T cells (ref. 9 of Appendix II). Tucaresol also exerts its effects on co-stimulation (ref. 10 of Appendix II). Because the co-stimulatory environment associated with conventional protein vaccines (and natural infections) is very different from the co-stimulatory environment associated with DNA vaccination, there would not be a reasonable expectation of success in using tucaresol with DNA vaccines and DNA immunization.

During 1995-96, large animal studies were indicating that adjuvants were likely to be needed for DNA vaccination and there was an expectation that, because of the fundamental differences between DNA vaccination and other forms of vaccination (e.g., using protein vaccines), adjuvants that worked for conventional protein vaccines would be unlikely to work for DNA vaccines (ref. 8 of Appendix II). It is for this reason that other approaches such as co-administering cytokines were pursued rather than adjuvants previously used for protein vaccines such as tucaresol (ref. 9 of Appendix II).

Another surprising feature of Applicants' claimed invention is the demonstration of amplification of the IgG1 subclass of antibody to mycobacterial HSP-65 by tucaresol when used in conjunction with a DNA vaccine. All previous observations of tucaresol have indicated that it switches immune responses to the Th1 type of response, amplifying Thi cytokines such as IFN-γ but not Th2 cytokines such as IL-4 (ref. 9 of Appendix II). However the IgG1 subtype is a Th2 subtype of antibody in the mouse.

Furthermore Applicants reiterate their comments made in the last response to the effect that known immunopotentiating agents have been tried in combination with DNA vaccines (as disclosed on page 3, lines 27-35, of the specification) with limited or mixed

success and the conventional adjuvants such as alum, FCA, and FIA are not effective as adjuvants in DNA vaccination, as demonstrated in Example 1 of Applicants' specification. A person of skill in the art would thus not have had a reasonable of success to assume that adjuvants successfully used in conventional protein vaccination would also be effective as adjuvants in DNA vaccination. It was all the more surprising therefore that Schiff base forming compounds such as tucaresol, an effective conventional vaccine adjuvant, could be successfully used in a DNA vaccine setting. The Examiner is therefore incorrect in citing Rhodes as providing a disclosure that alleges the usefulness of tucaresol in a DNA vaccine setting or even that there would be any motivation to use tucerasol in such a setting with any expectation of success.

The other reference cited by the Examiner, Herrmann et al., does not disclose any particular adjuvants or classes of adjuvants which might be expected to work, and provides no working examples using any such adjuvant. Moreover, the purpose of the adjuvant described by Herrmann et al. is to "promote DNA uptake" or "recruitment of immune system cells to the site." These might be termed adjuvants by Herrmann et al. but they are not the same as agents which would "enhance both humoral and cellular immune responses initiated by the antigenic peptide" as required by the claims of the present invention. This establishes that the term "adjuvant" is used to serve completely different purposes by Rhodes and Herrmann et al. The latter reference would not therefore provide any additional motivation to that disclosed in Rhodes, and in view of the lack of success reported with conventional adjuvants in enhancing the immune system's response to DNA vaccines, no expectation of success can be derived from the disclosure by Herrmann et al. of "adjuvants" which are intended to serve a completely different function.

The combined teachings of Rhodes and Herrmann et al. would not therefore suggest to a skilled artisan that Schiff base forming compounds such as tucaresol, will achieve enhancement of the immune response in a DNA vaccine setting, where other conventional adjuvants will not, nor that such compounds will achieve this utility by enhancing both the humoral and cellular immune responses initiated by the antigenic peptide expressed by the nucleotide sequence which forms the DNA vaccine. The

Bellhouse et al. reference was cited for its disclosure of a gene gun, which does not remedy the defects noted above with respect to Rhodes and Herrmann et al.

It was asserted on pages 4-5 of the Action that tucaresol provides (i) an immuno-potentiating response by directly engaging T cells and (ii) signals that converge with signal 1 resulting from TCR-antigen ligation (signal 1 is considered the same for natural infection, conventional vaccines, and DNA vaccines), but there was no evidence cited to support these assertions in the context of DNA vaccination. Similarly, it was asserted on page 5 of the Action that protein antigen can be secreted by transfected cells and then taken up by antigen presenting cells, but no evidence was presented to substantiate this assertion in the context of DNA vaccination. If these assertions are being relied upon by the Examiner, she is respectfully respected to provide evidence is support thereof.

Finally, Applicants note that new claim 25 is directed to enhancing "at least Th1 and Th2 associated responses" which is a limitation not taught or suggested by any of the cited references.

Withdrawal of the Section 103 rejection is requested because the invention would not have been obvious to a person of ordinary skill in the art at the time it was made.

Conclusion

Having fully responded to all of the pending objections and rejections contained in the Office Action (Paper No. 13), Applicants submit that the claims are in condition for allowance and earnestly solicit an early Notice to that effect. The Examiner is invited to contact the undersigned if any further information is required.

Respectfully submitted,

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